

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Serial No. 09/118,730
Filed: July 17, 1998
Inventors: Ellington M. Beavers et al
Title: METHOD OF MAKING FREE ACIDS
FROM POLYSACCHARIDE SALTS
Examiner: E. White
Art Unit: 1623
File No.: 281-28

SECOND DECLARATION OF ELLINGTON M. BEAVERS UNDER RULE 132

Ellington M. Beavers hereby declares as follows:

1. I am one of the named joint inventors in the above-identified application. I have read the Official Action of March 15, 1999, and I make this Declaration in response thereto.

2. At the time of filing of the present application, I submitted a Declaration under Rule 132, in which I described various experiments showing that the method used to make hyaluronic acid greatly affects the properties of the resulting product. I hereby incorporate that Declaration by reference herein. In my prior Declaration, I showed that a method disclosed in the prior art, for making hyaluronic acid, would not yield a product suitable for use in the body, due to its cytotoxicity, and due to its hemolytic tendencies. The conclusion of my prior Declaration, which I reiterate here, is that not all hyaluronic acid is alike. By the Amendment filed with this Declaration, I am now limiting the claims of this application to free hyaluronic acid which is suitable for use in the body. My prior Declaration shows that only hyaluronic acid made according to the present invention is suitable for use in the body.

3. Although the term "hyaluronic acid" appears in various references, that term has been loosely and imprecisely used, by professionals in the field, to refer either to hyaluronic acid or its salt. Indeed, many literature references use the term "hyaluronic acid" generically, even when what is really meant is its most common salt, namely sodium hyaluronate. Some references use the term "hyaluronic acid" when the context clearly indicates that only the salt was meant.

4. A very graphic example of the imprecise language described above is found in "Quantitative Approaches in the Histochemistry of Mucopolysaccharides", Journal of Histochemistry and Cytochemistry, vol. 11, pages 24-34 (1963). A copy of the latter article is attached as Exhibit A. In the explanation accompanying Figure 1, on page 26, the article describes various batches of sodium or potassium salts of mucopolysaccharides. Indeed, the substances described could not be free acids, because the text specifies a pH at which it is impossible for the acid form to exist. But while all of these substances are described as "salts", the hyaluronate is labeled "hyaluronic acid", while the other salts are correctly designated as "keratan sulfate" and "chondroitin sulfate". This terminology typifies the imprecise language that has been used for decades in the field. The term "hyaluronic acid" has been used, loosely, to refer to the polymer generically. It has been quite common to use the term "hyaluronic acid" when what is really meant is the sodium salt.

5. This confusion of terminology was placed in perspective in Laurent, "The Biology of Hyaluronan" (1989), which is a compilation of articles derived from an academic conference sponsored by the Ciba Founda-

tion. A copy of the Introduction of the latter reference is attached as Exhibit B. This document explains that at physiological pH, the carboxyl groups on the uronic acid residues are dissociated, and that the polysaccharide should be named "sodium hyaluronate" when sodium is the counter ion. Because it is often not possible to specify the counter ion, the reference proposes that the term "hyaluronan" be used to identify the polysaccharide in general terms. This convention would leave the terms "hyaluronic acid" and "hyaluronate" to refer to the free acid and salt forms of the polymer, respectively.

6. The proposal made in the Laurent reference has not been universally adopted; even today, the term "hyaluronic acid" is frequently used, incorrectly, to designate the sodium salt. Not only do academic references use the term "hyaluronic acid" inappropriately, but even various chemical companies still offer "hyaluronic acid" for sale, when only the sodium salt is really available for purchase.

7. Both prior to, and after, the making of the present invention, I or my associates in research have attempted to obtain small samples of free hyaluronic acid from commercial suppliers. I or my associates have inquired of all the major suppliers in the field, including Lifecore Biomedical, Sigma Chemical, Aldrich Chemical, Fluka Chemical, Acros Organics (Fisher Scientific), WMR Scientific Products, and Polysciences. In all but two cases, the suppliers indicated that they did not sell free hyaluronic acid, or that they could provide only the sodium salt. In one of the remaining two cases (Fluka), the supplier's catalog contained separate listings for various hyaluronic acid salts, as well as "hyaluronic acid from bovine vitreous humor". It thus appeared, at first, that the latter could

have been true hyaluronic acid. But when an inquiry was made to the technical department of the supplier, the supplier eventually admitted that the advertised product was actually the sodium salt, not the free acid form. That supplier later informed us that the supplier intended to correct the entry in its catalog. In the one remaining case, a supplier insisted that it could provide the free acid. But when we received the product and analyzed it, we determined that it was the sodium salt, not the free acid.

8. Thus, my own efforts have confirmed that free hyaluronic acid is not commercially available. Invariably, what is advertised as "hyaluronic acid" really proves to be, after analysis, the sodium salt. My experience thus shows that the mere mention of "hyaluronic acid", in any reference, must not be taken at face value, without proof that the substance is indeed the free acid.

9. The above conclusions can be applied to the references cited by the Examiner in the Official Action of March 15, 1999. Of the four references cited by the Examiner, only two (Schultz and de Belder) mention hyaluronic acid. I will discuss these below.

10. The patent to de Belder discloses a polysaccharide which, in every example except one, is described as sodium hyaluronate. The one exception occurs in Example 1(h), where the patent states that the free acid form of hyaluronic acid was used. However, the Example also states that the experiment was conducted "in the presence of 100 ul of glacial acetic acid". In all the examples in de Belder where sodium hyaluronate is used, glacial acetic acid is added to the reaction mixture in order to reduce the

pH to the range of 2 to 5 (preferably 2 to 4.5), as specified in the claims. But the pH of a solution of hyaluronic acid is already in the range of 2-5, as is disclosed in the present application, as well as in the corresponding parent patent (U.S. Patent No. 5,789,571). The pH of free hyaluronic acid would already be within the desired range, without the need to add glacial acetic acid. The only reasonable conclusion is that de Belder did not really use the free acid form, but instead used only the sodium salt. De Belder added glacial acetic acid because it was needed to reduce the pH of the sodium salt. It would not have been needed if the free acid form had been available.

11. The patent to Schultz (U.S. Patent No. 4,808,576) does recognize a distinction between free hyaluronic acid and its salt, and the patent purports to cover the use of the free acid. However, Schultz admits (column 4, line 61 through column 5, line 6) that all of the experiments described in the patent were performed with the sodium salt, not with the free acid. In fact, in column 5, lines 5-6, Schultz perpetuates the confusion described above, by explicitly defining the term "hyaluronic acid" to include both the free acid and the sodium salt. The Schultz patent therefore adopts the generic and misleading meaning of the term "hyaluronic acid", which meaning has been carelessly used by many others, for many years. But by its own admission, Schultz did not use or try the free acid form. Based on my own research described above, I doubt that Schultz had any source of free hyaluronic acid. Based on my attempts to locate the product, I believe that free hyaluronic acid would not have been available to Schultz, and that is why Schultz could not generate any data for free hyaluronic acid.

12. In summary, the references relied upon by the Examiner cannot be deemed to disclose free hyaluronic acid. Schultz admits that no free acid was used. And de Balder's own disclosure negates any inference that free acid was used.

13. Moreover, the references do not show free hyaluronic acid which is of a medical grade, i.e. which is suitable for use in the body. My prior Declaration showed that the method of manufacture determines whether the product is of medical grade, and that not all hyaluronic acid is alike. Thus, the failure of the references to disclose or suggest my method also means that the product made by my method is patentable.

14. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: July 9, 1969


Ellington M. Beavers